Laboratory Approval Program for Export of Meat and Poultry Products

1. Purpose

1.1 This document provides the requirements for the Laboratory Approval Program (LAP) for the Export of Meat and Poultry Products. This LAP is for laboratories seeking to perform confirmatory analysis of chemical residues, microorganisms, and parasites in meat and poultry products which are offered for certification by USDA Food Safety and Inspection Service (FSIS) for export to various countries.

1.2 It also provides the procedures and requirements used for objective evaluation of a laboratory's analysis program submitted for approval and monitored by the Agricultural Marketing Service (AMS), Science and Technology (S&T) Program, Laboratory Approval and Testing Division (LATD), Laboratory Approval Service (LAS).

2. Scope

This LAP may be used by laboratories that submit their analysis program to LAS for approval, verification, and monitoring. It is limited to the analysis of chemical residues, microorganisms, and T*richinella spiralis* in specified meat and poultry products and all aspects of a laboratory's documented quality management system that apply to these analyses.

3. References

3.1 USDA FSIS Export Library. FSIS webpage "Export Library – Requirements by Country" providing the access point of requirements for meat, poultry and processed egg products (http://www.fsis.usda.gov/wps/portal/fsis/topics/international-affairs/exporting-products/export-library-requirements-by-country).
3.2 EC 1984. Commission Directive of June 7, 1984 amending the Annexes to "Council Directive 77/96/EEC addresses the examination for trichinae (*Trichinella spiralis*) prior to importation from third countries of fresh meat derived from domestic swine."
3.3 EC 1976. Council Directive 77/96 EEC dated December 21, 1976. Official Journal of the European Communities. L 26: 67-77 and L 167:34-43.
3.4 EC 1994. Council Directive 77/96/EEC third amendment on December 2, 1994. Official Journal of the European Communities, L 315: 18-20.
3.5 EC 2002. Commission Decision of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results. Official Journal of the European Communities. L 221: 8-36.
3.6 EC 2004. Document SANCO 2726 rev 4 (December 4, 2008) Guidelines for the implementation of Decision 2002/657/EC (http://crl.fougeres.anses.fr/publicdoc/Guidelines-Consolidated_2002-657_2004-2726rev4_en.pdf).
3.7 EC 2007. Community Reference Laboratories for Residues CRL guidance paper, December 7 2007: CRLs view on state of the art analytical methods for national residue control

plans(http://crl.fougeres.anses.fr/publicdoc/2013/EURL_Guidance_Concentrations_Minimales_Recommend%C3%A9es_Methodes_Analytiques.pdf).

3.8 FDA 2006. Mass spectrometry for confirmation of the identity of animal drug residues. Center for Veterinary Medicine (CVM), Guidance for Industry #118. (http://www.fda.gov/downloads/AnimalVeterinary/GuidanceComplianceEnforcement/GuidanceforIndustry/UCM052658.pdf).

3.9 FDA 2011. Studies to evaluate the metabolism and residue kinetics of veterinary drugs in food-producing animals: Validation of analytical methods used in residue depletion studies. US FDA-VICH GL49. US FDA, Center for Veterinary Medicine, September 15, 2011. (http://www.fda.gov/downloads/AnimalVeterinary/GuidanceComplianceEnforcement/GuidanceforIndustry/UCM207942.pdf).

3.10 FDA 2012. Guidelines for the validation of chemical methods for the FDA Foods Program. US FDA, FDA Foods Program Science and Research Steering Committee, March 22, 2012. (http://www.fda.gov/downloads/ScienceResearch/FieldScience/UCM298730.pdf)

3.11 AOAC International Official Method, Appendix E: Laboratory Quality Assurance.

3.12 AOAC International Guidelines for Laboratories Performing Microbiological and Chemical Analyses of Food and Pharmaceuticals. Prepared by the Analytical Laboratory Accreditation Criteria Committee of AOAC International, revised March 2010.

3.13 40 CFR 792. EPA Good Laboratory Practice Standards http://www.ecfr.gov/cgi-bin/text-idx?SID=853d51742786f837131b5ad6e660f5b5&node=pt40.32.792&rgn=div5

3.14 USDA FSIS. Microbiology Laboratory Guidebook (MLG). http://www.fsis.usda.gov/wps/portal/fsis/topics/science/laboratories-and-procedures/guidebooks-and-methods/microbiology-laboratory-guidebook/microbiology-laboratory-guidebook

3.15 USDA FSIS. Chemistry Laboratory Guidebook (CLG). http://www.fsis.usda.gov/wps/portal/fsis/topics/science/laboratories-and-procedures/guidebooks-and-methods/chemistry-laboratory-guidebook.

4. Laboratory Approval Procedures

4.1 Initial Request for Admission: A laboratory seeking approval must send an email/letter to the Program Manager (PM) requesting admission to the program at the following address:

Program Manager – LAP-Export
Laboratory Approval & Testing Division
USDA, AMS, S&T
1400 Independence Ave SW
Room 3533-S
Washington DC 20250-0272
Telephone: (202) 690-0621
Email: LAS@ams.usda.gov

4.2 Submission of Required Information: After providing the initial request for admission into the program, the applicant laboratory must address all program requirements, including fees (see Section 10). The following required materials must be provided to the PM for review:

4.2.1 A letter from the management providing information as to:

4.2.1.1 corporate entity, name, address, phone number, email address, and legal status;

4.2.1.2 a clearly defined scope for the approval;

4.2.1.3 the name(s), title(s) and contact information of laboratory staff designated to serve as Approved Signatory(ies) of test reports that reference USDA-approved laboratory;

4.2.1.4 conflict of interest statement; and

4.2.1.5 signature of an authorized representative of the applicant laboratory.

4.2.2 The laboratory must be ISO/IEC 17025 accredited. The methodology used for the LAP must be a part of the scope of accreditation. The laboratory must provide up-to-date copies of the scope of accreditation. Note: Laboratories providing trichinae analysis may choose to opt out of being ISO/IEC 17025 accreditation since these laboratories are overseen by the USDA FSIS on-site inspectors.

4.2.3 A description of the physical condition of the laboratory and the capabilities in terms of major and critical equipment/instruments for the analysis.

4.2.4 An organization chart showing the overall laboratory structure. Additional documents detailing the relevant person's (quality manager and analysts at a minimum) laboratory functions, qualifications and training procedures/records.

4.2.5 Standard operating procedures (SOPs) – These include, but are not limited to, sample receiving and inspection (weight, seal, and sample condition), the analytical methods, quality assurance and quality control, instrument calibration, reporting test results, and equipment maintenance.

4.2.6 Method validation– The laboratory must submit method validation data and results (See Section 11).

4.2.6.1 Laboratories are required to participate in an external ISO 17043 accredited proficiency testing (PT) program(s) when available. Analyses of Certified References Materials (CRMs), where available and applicable, must be included in the validation process.

4.2.6.1.1 A PT program for trichinae analysis will be administered by AMS. AMS will contract with the USDA Agricultural Research Service to prepare PT samples and ship samples to the laboratories. The cost for this service is detailed in section 10.5.

4.2.6.2 Laboratories must send PT results to the PM (and reports of corrective actions if the PT results indicate insufficient performances).

4.2.7 The program is user-fee supported and all laboratories must pay program fees upon receipt of the billing invoice. All fees must be received prior to admission to the program (See Section 10).

4.2.8 For billing purpose, the laboratory needs to provide Federal W-9 Form showing the laboratory's taxpayer identification number, or Federal W-8BEN Form (Certificate of Foreign Status of Beneficial Owner for United States Tax Withholding) showing the

"Foreign tax identifying number" of a foreign laboratory. The laboratory also needs to supply the contact person(s) of account payable.

4.3 Review of Information Submitted: The PM will review the required materials and communicate to the applicant laboratory any concerns or deficiencies. The applicant laboratory must respond, in writing, to the concerns or deficiencies.
4.4 Pre-admission Sample Testing: The PM may request additional analytical results from the laboratory for sufficiently evaluating the laboratory's capability for the program at the laboratory's cost. These additional materials could result from analysis of internal QC samples, and/or external QC samples.
4.5 Initial Onsite Laboratory Audit: LATD personnel will conduct an onsite laboratory audit after the above conditions have been met successfully.
4.6 Issuance of Acceptance Letter: AMS will provide a letter of approval to the applicant laboratory after it meets all program requirements.
4.7 Approved laboratory will be added to the official list of participating laboratories which is maintained and published on the LATD website.

NOTE: The approval expires at the end of year and is renewable on January 1st of every following year.

5. Maintaining Program Status

5.1 The LAP is managed on a calendar year basis (January 1 – December 31 of each year).
5.2 Laboratories must maintain their ISO/IEC 17025 accreditation. The methodology used for the LAP must be a part of the scope of accreditation and laboratories must provide up-to-date copies of the certificate and the scope of accreditation. Note given in 4.2.2 applies here to laboratories providing trichinae analysis.
5.3 Laboratories must analyze samples administered by external ISO 17043 accredited PT institution(s), when available, and meet a satisfactory status in terms of Z-score, recovery ratio, and consistency. Laboratories send PT results to the PM (and reports of corrective actions if the PT results indicate insufficient performances). Item 4.2.6.1.1 applies here to laboratories providing trichinae analysis.
  5.3.1 For analysis where no external PT program is available, laboratories verify their methods annually by conducting internal verification process and by following relevant ISO standards or guidelines as given in the above references.
  5.3.2 Overtime, every analyst performing the method(s) must participate in the validation process and external PT program and submit the results with analyst name to the PM.
5.4 Only with the PM's approval, laboratories may change their operations. Those operations include but are not limited to: location, SOPs, equipment, instruments, and analysts. Any change may require the laboratory to verify or valid the analytical method.
  5.4.1 Upon change of analyst, the laboratory must inform the PM with the training record and the results of method verification study performed by the new analyst.
  5.4.2 The laboratory must inform the PM if its name, physical location, or contact information has changed.

5.4.3 Not informing the PM of these changes may lead to immediate removal from this program which would require the laboratory to re-apply in order to participate in the program.

5.5 All information relevant to the program must be made available to the PM upon request.

5.6 A laboratory is audited on-site once every two years. The laboratory must pass the onsite audit or make corrective actions in a timely manner.

5.7 At any time, if there is concern about the laboratory's ability to meet program requirements, AMS may conduct an additional onsite audit of a laboratory at the laboratory's expense.

5.8 The laboratory has paid the program fee in full.

6. Removal from the Program

6.1 Voluntary Removal: A laboratory may voluntarily remove itself from the program at any time by submitting a written request to the PM.

6.2 Involuntary Removal: A laboratory may be involuntarily removed from the approval program with any one of the following activities, but are not limited to:

6.2.1 Falsification of analytical results.

6.2.2 Failure to use methods and procedures approved by AMS.

6.2.3 Failure to meet analytical requirements.

6.2.4 Failure to maintain an acceptable performance level as indicated by the results of PT and/or internal verification samples.

6.2.5 Failure to perform corrective actions to address non-conformances in a timely and/or acceptable manner according to the PM and/or the auditor(s).

NOTE: The yearly program fee will not be refunded (whole or prorated), regardless of voluntary removal or involuntary removal.

7. Readmission to the Program

7.1 A laboratory that is removed from the program due to falsification of analytical results will be prohibited from reapplying.

7.2 A laboratory involuntarily removed from the program must wait, at least, for six months before it can re-initiate the admission process.

7.3 A voluntarily removed laboratory may be admitted back to the program within one program year (one calendar year) without paying additional fees, on the condition that no changes have occurred since the voluntarily removal in terms of equipment, instruments, analysts, methods, PT program participation, etc.

7.4 A laboratory voluntarily removed in the previous program year may be admitted back to the program during the current program year by paying the yearly program fee (See Section 10), on the condition that the time lapse from the voluntarily removal is less than one year (365 days), and no changes have occurred since the voluntarily removal in terms of equipment, instruments, analysts, methods, PT program participation, etc.

8. Complaints

All complaints should be brought to the attention of the PM for timely resolution. If the complaint cannot be resolved by the PM to the satisfaction of the complainant, the complaint may be brought to the attention of the Director of the LATD. The contact information for the current Director is as follows:

Kerry R. Smith, Ph.D., Director
Laboratory Approval & Testing Division
USDA, AMS, S&T
1400 Independence Ave. SW
Room 3533-S
Washington, D.C. 20250-0272
Telephone: (202) 690-4089
Email: KerryR.Smith@ams.usda.gov

9. Appeals

9.1 A laboratory that has been involuntarily removed from the program may file a written appeal to the Deputy Administrator of the S&T Program with supporting evidence as to why the laboratory should not be removed from the program. Within 30 days of receipt of the written appeal, the Deputy Administrator shall make a determination and take action, as deemed appropriate, with respect to the removal. The name, address, and telephone number of the current Deputy Administrator are as follows:

Ruihong Guo, Ph.D., Deputy Administrator
USDA, AMS, Science and Technology
1400 Independence Ave. SW, Room 3543-S
Washington, D.C. 20250-0270
Telephone: (202) 720-8556
Email: Ruihong.Guo@ams.usda.gov

9.2 If the appeal to the Deputy Administrator of S&T Program cannot be resolved to the satisfaction of a laboratory, an appeal, in writing, may be filed with the Administrator of AMS. Within 90 days of receipt of the written appeal with supporting evidences, the Administrator shall make a determination and take action, as deemed appropriate, with respect to the removal. The name, address, and telephone number of the current Administrator are as follows:

Anne Alonzo, Administrator
USDA, Agricultural Marketing Service
1400 Independence Ave. SW, Room 3071-S
Washington, D.C. 20250-0201
Telephone: (202) 720-4276

10. Fee Schedule

10.1 LATD sets the program fees (e.g., admission fee, initial yearly fee, and yearly fee) for each program based on administrative costs. Program fees are reviewed yearly to determine if they are adequate compared with USDA operational costs.

10.2 All program fees are neither refundable nor prorated.

10.3 Program Fees

- 10.3.1 The admission fee covers the cost of initial management: reviewing application materials, initial laboratory evaluation, follow-up communication, and account set up.
- 10.3.2 The initial yearly fee covers the initial onsite audit, and first year on-going management if the laboratory is accepted into the program.
- 10.3.3 If the initial on-site audit cannot be started within a year (365 days starting from the applicant's submission day of required information) due to the applicant's delinquency or incapability, the applicant needs to pay the admission fee again in the following year to keep the admission process.
- 10.3.4 The yearly fee covers the on-going management cost, including on-site audit, after a laboratory has been accepted into the program.

10.4 The LAP-Export program covers several groups of analytes, as listed below. One laboratory may provide analytical services for one or a combination of several groups:

- 10.4.1 Chemical residues
  - 10.4.1.1 AB: Antibiotics: tetracyclines and chloramphenicol
  - 10.4.1.2 BA: Beta agonists: ractopamine, clenbuterol, and zilpaterol
  - 10.4.1.3 HM: Heavy metals: arsenic, lead, cadmium and mercury
  - 10.4.1.4 PC: Pesticides: DDT, dieldrin, and lindane
  - 10.4.1.5 RC: Resorcylic acid lactones: taleranol and zeranol
  - 10.4.1.6 SR: Steroids: melengestrol acetate and trenbolone
- 10.4.2 Microorganisms
  - 10.4.2.1 LM: *Listeria monocytogenes*
  - 10.4.2.2 SM: *Salmonella*
  - 10.4.2.3 TPC: Total plate count (microorganisms per gram or mL)
- 10.4.3 Parasites
  - 10.4.3.1 TS: Trichinae: *Trichinella spiralis*

10.5 The program fee is based on the number of groups for which a laboratory offers to provide analytical services. The calculation of fees is illustrated in the following examples.

- 10.5.1 A laboratory selects to analyze 5 groups of chemical residues. The admission fee, initial yearly fee and yearly fee are $2690, $6180, and $3970, respectively.
- 10.5.2 A laboratory selects to analyze 2 groups of microorganisms and 2 groups of chemical residues. The three fees are $2230, $5590, and $3610, respectively.
- 10.5.3 The on-site training fee for analyzing trichinae is $3120.
- 10.5.4 If international travel is required for an on-site audit, an additional fee will be charged to the laboratory. Contact the program manager for details.

# of Groups	Admission Fee ($)	Initial Yearly Fee ($)	Yearly fee ($)	Trichinae Training Fee ($)	Trichinae PT Program per Analyst ($)
1	870	3840	2600	3120	1750
2	1320	4420	2930		
3	1780	5010	3250		
4	2230	5590	3610		
5	2690	6180	3970		
6	3140	6760	4320		
7	3600	7350	4680		
8	4050	7930	5040		
9	4510	8520	5400		
10	4960	9100	5750		

11. Technical Requirements

11.1 Chemical Analysis

11.1.1 Methods

11.1.1.1. Participant laboratories must use LATD specified and/or LATD accepted methods.

11.1.1.2. If methods are not specified, LATD allows laboratories to use methods from different sources such as AOAC International Official Methods, US FDA methods, USDA FSIS methods, or US EPA methods. Laboratories may also adopt methods which are published/used by other national or international organizations.

11.1.1.3. If methods from the above sources are not available or not very relevant, laboratories can adapt methods from those sources into a laboratory method.

11.1.1.4. Methods from different sources must be relevant to the program in terms of matrices, concentration ranges, and methodology.

11.1.1.5. Semi-developed methods, in-house methods, proprietary methods without details being accessible by the PM, and similar methods are in general not permitted.

11.1.2 Standard Operating Procedures (SOP)

11.1.2.1. Laboratories must establish project-oriented and program-specific SOPs.

11.1.2.2. These SOPs must be operational under specific laboratory conditions (in terms of facilities and operator's qualification).

11.1.2.3. These SOPs must be reviewed and signed by quality managers, laboratory directors and/or by people with similar responsibilities.

11.1.2.4. These SOPs must be periodically reviewed (at least once every two years) and updated to the latest standards.

11.1.2.5. Manufacturer's Instrument Instructions/Manuals and similar documents are not accepted as substitutions for laboratory SOPs.

11.1.3 Analytes, Sensitivities, Method Detection Limits, and Specified Methods
Background: LATD manages this LAP-Export at the request of USDA FSIS so that establishments have means to ensure meat and poultry products comply with the FSIS Export Library. The Export Library declares sensitivities which are treated as maximum residue limits (MRL or MRLs) of analytes for export purpose. These sensitivities may be close to method detection limits (MDL or MDLs, such as 0.1 ppb of ractopamine in meat) or much higher than method detection limits of currently available methods (such as 500 ppb of lead in meat). Laboratories must be capable so that residues at or below these sensitivities are reliably detected and measured. Therefore, MDL, an indication of a laboratory's analytical capability, may be required to be lower than these sensitivities, or may be allowed to be close to these sensitivities, depending on sample matrices, residue concentration ranges, analytical challenges, other program requirements, etc.

The residues are divided into groups. Each group may contain several analytes (Note: µg/g = µg/mL = ppm, µg/kg = µg/L = ppb).

11.1.3.1 Antibiotics
11.1.3.1.1. Methods: LC-MS/MS
11.1.3.1.2. Matrix: Liver, muscle
11.1.3.1.3. List of analytes

Analyte	Sensitivity (ppb)	MDL (ppb)
Tetracycline	10	≤ 10
Chlortetracycline	10	≤ 10
Oxytetracycline	10	≤ 10
Chloramphenicol	10	≤ 10

11.1.3.2 Beta agonists
11.1.3.2.1 Methods: LC-MS/MS
11.1.3.2.2 Matrix: Liver, muscle
11.1.3.2.3 List of analytes

Analyte	Sensitivity (ppb)	MDL (ppb)
Ractopamine	liver 0.2, muscle 0.1	≤ 0.1
Clenbuterol	liver 0.2, muscle 0.1	≤ 0.1
Zilpaterol	liver 1, muscle 1	≤ 0.5

11.1.3.3 Heavy Metals
11.1.3.3.1 Methods: USDA FSIS CLG-TM3. Determination of metals by ICP-MS and ICP-OES (Rev: 04, 09/30/2013) or equivalent

11.1.3.3.2 Matrix: Muscle
11.1.3.3.3 List of analytes

Analyte	Sensitivity (ppb)	MDL (ppb)
Arsenic (As)	100	≤ 10
Cadmium (Cd)	50	≤ 10
Lead (Pb)	500	≤ 25
Mercury (Hg)	30	≤ 20

11.1.3.4 Pesticides
11.1.3.4.1 Methods: USDA FSIS CLG-CHC3. Determination of chlorinated hydrocarbons (CHCs) and chlorinated organophosphate hydrocarbons (COPs) with gel permeation chromatography (GPC) (Rev: 04, 06/01/2010) or equivalent. AOAC International Official Method 970.52, Organochlorine and organophosphorus pesticide residues or equivalent
11.1.3.4.2 Matrix: Fat
11.1.3.4.3 List of analytes

Analyte	Sensitivity (ppb)	MDL (ppb)
DDT and its metabolites	100	≤ 50
Dieldrin	300	≤ 50
Lindane (γ-HCH)	100	≤ 50

11.1.3.5 Resorcylic acid lactones
11.1.3.5.1 Methods: LC-MS/MS
11.1.3.5.2 Matrix: Liver, muscle, urine
11.1.3.5.3 List of analytes

Analyte	Sensitivity (ppb)	MDL (ppb)
Taleranol	1	≤ 1
Zeranol	1	≤ 1

11.1.3.6 Steroids
11.1.3.6.1 Methods: LC-MS/MS
11.1.3.6.2 Matrix: Liver, muscle, urine
11.1.3.6.3 List of analytes

Analyte	Sensitivity (ppb)	MDL (ppb)
Melengestrol acetate	Liver 5, muscle 1	≤ 1
Trenbolone	Liver 5, muscle 1	≤ 1

11.1.3.7 Other analytes not listed above:

AMS approved laboratories may analyze other infrequent residues on the condition that the analytical methods are on the scope of the laboratory's ISO/IEC 17025 accreditation.

11.1.4 Calibration

11.1.4.1 Calibration range: Typical concentrations are set at a minimum of 5 different levels below and above the sensitivities. The calibration range, expressed as analytes in sample, should cover from ≤ 50% of MDL to ≥ 2-10 × sensitivity.

11.1.4.2 Matrix effect: Laboratory must evaluate the specific and non-specific interferences caused by sample matrix, with respect to clean solvent/buffer. Laboratory must evaluate the effect of applying internal reference standard, matrix-matching, internal standard addition, application of stable isotopes, etc., for relieving or eliminating matrix effect. Evidence must be provided to the PM to indicate the matrix effect is corrected to a satisfactory degree.

11.1.4.3 Calibration matrix: Calibrators are prepared in a minimum of two types of matrix: Type 1 – Standards in clean solvent/buffer; and Type 2 – Standards fortified into control matrix extract (tissue extract). The two responses (as slopes of their calibration lines) are within 100±30% of each other, in general. Laboratory must decide the most optimum calibration for unknown samples

NOTE: "Control matrix", "Negative control", or "Blank matrix", is the animal tissue which is confirmed to be chemical residue free based the known history of animals which have not been exposed to regulated or forbidden drug/chemicals, or based on previous measurements. "Control matrix extract" is the extracted solution after the control matrix is processed through the whole procedure but the measuring process. "Solvent/buffer" is the solution which is used to prepare the control matrix extract right before the measuring process.

11.1.5 Selectivity (Specificity) required in LC-MS/MS analysis, when applicable.

11.1.5.1 Deuterated standards shall be used as an internal reference standard when applicable.

11.1.5.2 Chromatographic separation: The drug residue retention time of a sample shall match that of calibration standard within an accepted window.

11.1.5.3 The retention time ratio of drug residue to deuterated standard of a sample shall correspond to that of calibration standard at a tolerance of ±2.5%.

11.1.5.4 The relative intensities of fragment ions of a sample shall match that of the calibration standard.

11.1.5.5 At least one precursor and two daughter ions shall be identified

11.1.6 Sensitivity (in terms of limit of detection and limit of quantitation)

11.1.6.1 Establishing limit of detection (LOD)

11.1.6.1.1 Analyze at least 12-20 blank samples (tissue extract from blank samples, n > 12).

11.1.6.1.2 Convert the noise signal to concentration at the time window in which drug residue is expected.

11.1.6.1.3 Calculate the average (avg) of all those (noise) concentrations and the standard deviation (sd).

11.1.6.1.4 Let P = 3 × avg.

11.1.6.1.5 Let Q = avg + 1.64 × sd.

11.1.6.1.6 LOD is the greater value out of P and Q.

11.1.6.2 Limit of quantitation (LOQ): LOQ is the mean plus 10 standard deviations of the above mentioned measurements

11.1.7 Sensitivity (in terms of method detection limit and method quantitation limit) The above LOD and LOQ should be converted to method detection limit (MDL) and method quantitation limit (MQL), accordingly, such as by applying proper dilution factors. These MDL and MQL shall be expressed as drug residue in tissue.

11.1.8 Accuracy – It is the closeness (trueness) of measured concentration to confirmed concentration of an analyte in a certified reference material (CRM) containing incurred analyte. Trueness may be calculated as bias (= 100% × ($C_{Measured}$ - $C_{Certified}$)/ $C_{Certified}$)) and the requirements are listed below

Concentration (ppb)	Range of Trueness (%) (n > 6)
≤ 1 (e.g. 0.3)	-50 - +20
1 - 10 (e.g. 3)	-40 - +10
10 - 100	-30 - +10
> 100	-20 - +10

NOTE: According to Commission Decision 2002/657/EC (EC 2002), when no such CRM is available, it is acceptable that trueness of measurements is assessed through recovery of additions of known amounts of the analyte(s) to a blank matrix. Data corrected with the mean recovery are acceptable only when they fall within the ranges shown above.

When trueness is expressed and calculated as recovery (100% × $C_{Measured}$ / $C_{Certified}$), the requirement is given below in the "Recovery" section

11.1.9 Precision – It is evaluated by fortifying chemical standards in control matrix at 1× and 3× the sensitivity levels as given in Section 11.3 (0.5 and 3 ppb if the required sensitivity of 11.3 is 0.1ppb). The acceptable precision (coefficient of variation, CV = 100% × one standard deviation / mean of repeated measurements) is listed below

Analyte concentration (ppb)	Within-run precision (Repeatability) (%CV) (n > 10)	Between-run precision (Reproducibility) (%CV) (n > 10)
≤1	30	45
1 - 10	25	32
10 - 100	20	23
100-1000	15	20
> 1000	10	16

11.1.10 Recovery

Analytes are fortified to control matrix (tissue) at 1× and 3× the sensitivity levels as given in Section 11.3 (0.5 and 3 ppb if the required sensitivity of 11.3 is 0.1ppb). The recovery (= 100% × $C_{Measured}$ / $C_{Fortified}$) of fortified chemical standards meets the following requirements.

Fortified concentration (ppb)	Recovery (%) (n > 6)
≤ 1	50 - 120
1 - 10	60 - 110
10 - 100	70 - 110
> 100	80 - 110

11.1.11 Extension of Sample matrix

A method validated on one sample matrix (e.g. pork muscle) is extended to other sample matrix (e.g. turkey muscle) in the following general ways. Six sets for each species/matrix combination (e.g. heavy metals in turkey muscle); each set consist of six samples: two blanks, two samples at sensitivity level, one sample at 2× sensitivity level, and one sample at 4 × sensitivity level. Two sets of samples are analyzed in one day and six sets of samples are analyzed in three different days. The results must meet the above specifications.

11.2 Microbiological Analysis

11.2.1 Microorganisms, Sensitivity, and Methods

11.2.1.1 *Salmonella*

11.2.1.1.1 Methods: AOAC International Official Method, US FDA methods, and/or USDA FSIS methods that specifies: All Foods; Raw, highly contaminated foods; or Poultry; and tests for all *Salmonella* (both motile and non-motile).

11.2.1.1.2 Sensitivity: 0 (negative in 25 gram of tested sample).

11.2.1.2 *Listeria monocytogenes*

11.2.1.2.1 Methods: AOAC International Official Method, US FDA Bacteriological Analytical Manual (BAM), and/or USDA FSIS method may be used.

11.2.1.2.2 Sensitivity: 0 (negative in 25 gram of tested sample).

11.2.1.3 Total Plate Count

11.2.1.3.1 Methods: AOAC International Official Methods (Petrifilm Plate count Method), US FDA methods, USDA FSIS methods, and/or Compendium of Methods for the Microbiological Examination of Foods.

11.2.1.3.2 Sensitivity: 10 CFU/g (colony forming units/g).

11.2.2 Other Methods

Other methods (such as rapid screening methods) used to test for pathogenic microorganisms must have been tested against a reference cultural method.

NOTE: The reference method is defined as that method by which the performance of an alternate method is measured or evaluated. Validation studies must include comparison to a recognized reference method to demonstrate equivalence or increased performance, the significance of which must be determined statistically. For bacterial analytes, reference methods are generally culture-based and result in a pure isolate. The AOAC International Official Methods, the US Food and Drug Administration, Bacteriological Analytical Manual (BAM), the USDA, Food Safety and Inspection Service, Microbiology Laboratory Guidebook (MLG) and International Standards Organization (ISO) all contain culture methods that are recognized reference culture methods.

A laboratory using other methods must conduct a specific validation of those methods against reference culture method (s) to validate inclusivity, exclusivity, sensitivity and the methods performance as established by collaborative study. When new rapid methods based on DNA test technology are used, method accuracy, precision, repeatability, and reproducibility should also be considered.

The validation should challenge the methods ability to detect the pathogen of interest in samples inoculated with low and high levels concentrations of the target organism, as-well-as samples inoculated with competitive levels of other organisms including the test organism. One should also consider samples that are naturally contaminated as well as samples inoculated with concentrations of competing organism at levels higher than that of the target organism. The results from these samples should demonstrate very low or no false positive/false negative rates.

Any samples tested positive by those other methods must be confirmed by reference culture methods. Cultural confirmation includes the use of biochemical and serological tests to demonstrate that the other method did properly detect the targeted test organism.

11.3 Trichinae Analysis

11.3.1 Location: The laboratory must be located onsite where the product is processed.

11.3.2 Analyst Training: Analysts must be trained by AMS to receive certification.

11.3.2.1 The training will consist of both lecture (including test method) and laboratory.

11.3.2.2 The training will occur onsite at the laboratory.

11.3.2.3 Upon completion of the training, the analysts must successfully analyze an initial or second set of initial proficiency samples in his/her own laboratory to complete his/her certification.

11.3.3 The company/establishment and laboratory must conform to the Council Directive 77/96/EEC of 21 December 1976 on the examination of *Trichinae* (*Trichinella Spiralis*) upon importation from third countries of fresh meat derived from domestic swine.

11.3.4 Test Method

11.3.4.1 The following acid digestion methods are accepted into this program: Magnetic Stirrer Method for Pooled Sample Digestion (Magnetic Stirrer method) and Mechanically Assisted Pooled Sample Digestion Method, Sedimentation Technique (Stomacher method).

11.3.4.2 The laboratory can use either one or both of the above methods to conduct the analysis.

NOTE: The Magnetic Stirrer method is included in the Trichinella Analyst Training Manual, which is provided in the training session.